λ CHAINS
Turbidimetric Method

Determination of Free and Bound Lambda Chains in Serum and Plasma

2 + 20 ml **REF 4561**

Additional required reagent:
λ chains calibrator (1 x 1 ml) **REF 4601**

PRINCIPLE

Antisera are a mixture of specific antibodies obtained by immunizing guest animals (goat) with human proteins. Titre, avidity and purity of these antibodies make them suitable to be used in turbidimetry. In this kind of analysis, the serum proteins present in the sample react immunologically with antiserum (anti-protein) to form a turbid reticulate which can be measured by photometer.
The measure, correlated to a calibration curve, is proportional to the concentration in the sample.

REAGENTS

Kit components: **REF 4561**

REAGENT 1 Antisera activator (liquid) **2 x 10 ml**
Buffered solution with PEG, stabilizers and preservatives.
Ready for use.

REAGENT 2 Antisera (liquid) **2 x1 ml**
Antiserum: researched anti-protein, in buffered solution.
Ready for use.

REAGENT 3 Samples and calibrators diluent (liquid) **2 x 10 ml**
Saline solution with stabilizers and preservatives. Ready for use.

WARNING: The reagents contain sodium azide (< 0.1%). Handle with care. Do not swallow. Avoid contact with skin and mucous membranes.

STABILITY: stable at 2-8°C up to the expiration date on the label.

REQUIRED EQUIPMENT

Spectrophotometer or filter photometer (340 nm).

SAMPLE

Serum or plasma.
STABILITY: 4 days at 2-8°C, 4 weeks at -20°C.

PREPARATION OF THE REAGENTS AND SAMPLES

Bring reagents and samples to room temperature before use.
Use Reagent 3 to dilute 1:20 samples and calibrators.
Draw the calibration curve for each test.

PREPARATION OF THE CALIBRATION CURVE

In order to draw the calibration curve, dilute k and λ chains Calibrator (Code 4601) with Reagent 3 to obtain 5 concentration levels included between the extremes of linearity range.
The procedure and the dilutions are explained in the calibrator instruction sheet. Always use 5 concentration levels to obtain an optimum calibration.

LINEARITY

150 – 1000 mg/dl.

MANUAL ASSAY PROCEDURE

Wavelength:	340 nm
Optical path:	1 cm
Reading:	against blank sample
Temperature:	30-37°C
Method:	endpoint
Reaction time:	10 minutes
Sample/R1/R2:	see table below

Label a number of cuvettes for the samples and 5 cuvettes for the five calibration levels.
Then proceed according to the following scheme:

	Sample	Calibrators
Reagent 1	500 µl	500 µl
Samples (diluited 1:20)	25 µl	----
Calibrators (diluited 1:20)	---	25 µl
Mix and measure A1 absorbances		
Reagent 2	50 µl	50 µl
Mix, wait 10 minutes and measure A2 absorbances		

CALCULATION

1. Draw a Cartesian system on graph paper: Concentration should be along the horizontal (X) axis as the (mg/dL) and the absorbance along the vertical (Y) axis.
2. Mark Δ absorbances (A2-A1) of the 5 calibrator levels in comparison with the corresponding concentrations.
3. Draw a line of best of fit through the five points.
4. Mark Δ absorbances (A2-A1) relative to the samples and refer to the researched concentrations using the curve.

REFERENCE VALUES

λ chains: 330-720 mg/dl.
Every laboratory should determine its own reference range.

NOTES

1. (*) Dangerous reagents are marked by an asterisk. Refer to MSDS.
2. A proportional variation in reagent volumes does not change results.
3. The concentration values stated in this instruction sheet refer to the weight of the whole protein (light fraction + heavy fraction). Divide by 3 to obtain the reference value of light fraction only.

REFERENCE

Available upon request.

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